Applicant: Franklin R. Cockerill III et al. Attorney's Docket No.: 20014-0004002 / 22515 US2

Serial No.: 10/754,223 Filed: January 9, 2004

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Amendments to the Specification:

Please amend the paragraph beginning at page 20, line 1, of the specification as follows: Conventional PCR methods in conjunction with FRET technology can be used to practice the methods of the invention. In one embodiment, a LightCyclerTM instrument is used. A detailed description of the LightCycler™ System and real-time and on-line monitoring of PCR can be found at http://biochem.roche.com/lightcycler on the World Wide Web. The following patent applications describe real-time PCR as used in the LightCyclerTM technology: WO 97/46707, WO 97/46714 and WO 97/46712. The LightCycler™ instrument is a rapid thermocycler combined with a microvolume fluorometer utilizing high quality optics. This rapid thermocycling technique uses thin glass cuvettes as reaction vessels. Heating and cooling of the reaction chamber are controlled by alternating heated and ambient air. Due to the low mass of air and the high ratio of surface area to volume of the cuvettes, very rapid temperature exchange rates can be achieved within the LightCyclerTM thermal chamber. Addition of selected fluorescent dyes to the reaction components allows the PCR to be monitored in real-time and online. Furthermore, the cuvettes serve as an optical element for signal collection (similar to glass fiber optics), concentrating the signal at the tip of the cuvettes. The effect is efficient illumination and fluorescent monitoring of microvolume samples.

Please amend the paragraph beginning at page 21, line 22, of the specification as follows: Another FRET technology format utilizes TaqMan® technology to detect the presence or absence of an amplification product, and hence, the presence or absence of *B. pertussis* or *B. parapertussis*. TaqMan® technology utilizes one single-stranded hybridization probe labeled with two fluorescent moieties. When a first fluorescent moiety is excited with light of a suitable wavelength, the absorbed energy is transferred to a second fluorescent moiety according to the principles of FRET. The second fluorescent moiety is generally a quencher molecule. During the annealing step of the PCR reaction, the labeled hybridization probe binds to the target DNA (i.e., the amplification product) and is degraded by the 5' to 3' exonuclease activity of the Taq Polymerase during the subsequent elongation phase. As a result, the excited fluorescent moiety and the quencher moiety become spatially separated from one another. As a consequence, upon excitation of the first fluorescent moiety in the absence of the quencher, the fluorescence

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emission from the first fluorescent moiety can be detected. By way of example, an ABI PRISM® 7700 Sequence Detection System (Applied Biosystems, Foster City, Calif.) uses TaqMan® technology, and is suitable for performing the methods described herein for detecting *Bordetella*. Information on PCR amplification and detection using an ABI PRISM® 770 system can be found at http://www-appliedbiosystems.com/products on the World Wide Web.

Please amend the paragraph beginning at page 26, line 8, of the specification as follows: Probes were obtained from Idaho Technologies (<u>available at http://www.idahotech.com/itbiochem/index.html on the World Wide Web</u>). The probes were suspended in 1X TE buffer supplied with the probes to a final concentration of 20 µM.